510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
DEVICE ONLY TEMPLATE

A. 510(k) Number:
k042519

B. Purpose for Submission:
New device

C. Analyte:
1,25-Dihydroxy Vitamin D

D. Type of Test:
Radioimmunoassay

E. Applicant:
Immunodiagnostic Systems, Ltd.

F. Proprietary and Established Names:
Gamma-B 1,25-Dihydroxy Vitamin D RIA

G. Regulatory Information:

1. Regulation section:
   21 CFR § 862.1825, Vitamin D test system

2. Classification:
   Class II

3. Product Code:
   MRG, Vitamin D test system

4. Panel:
   Chemistry (75)

H. Intended Use:

1. Intended use(s):
   See Indications for Use

2. Indication(s) for use:
   The IDS Gamma-B 1,25-Dihydroxy Vitamin D kit is a complete assay system intended for purification of 1,25-dihydroxyvitamin D (1,25D) in human serum or plasma by immunoextraction followed by quantitation by ^{125}I radioimmunoassay.

Results are to be used in conjunction with other clinical and laboratory data to assist the clinician in the assessment of 1,25D deficiency associated with renal disease.

3. Special condition for use statement(s):
   For in vitro diagnostic use only. Results are to be used in conjunction with other clinical and laboratory data.

4. Special instrument Requirements:
   Instrumentation common to most high complexity laboratories: centrifuge, sample mixer, vortex, gamma counter

I. Device Description:

The IDS Gamma-B 1,5-Dihydroxy Vitamin D kit is a complete assay system of the purification of 1,25D in patient samples by immunoextraction followed by quantitation by ^{125}I RIA. Patient samples are dilapidated and 1,25D extracted from potential cross-reactants by incubation for 3 hours with a highly specific solid phase monoclonal anti-1,25D. The immunoextraction gel is then washed and purified 1,25D eluted directly into glass assay tubes. Reconstituted eluates and calibrators are incubated overnight with a highly specific sheep anti-1,25D. ^{125}I-1,25D is added and incubation continued for 2 hours. Separation of bound from free is achieved by a short incubation with Ac-Cel® followed by centrifugation, decantation and counting. Bound radioactivity is inversely proportional to the concentration of 1,25D.

Controls included in the device are prepared from human source materials. They have tested negative by FDA recommended assays for the presence of antibodies to HIV (I and II), to HCV, and to Hepatitis B surface antigen.

J. Substantial Equivalence Information:

1. Predicate device name(s):
   DiaSorin 1,25-Dihydroxyvitamin D ^{125}I RIA

2. Predicate K number(s):
   k014030

3. Comparison with predicate:

Similarities		
Item	**Device**	**Predicate**
Intended Use	Quantitative determination of 1,25D for assessment of deficiency associated with renal disease	Quantitative determination of 1,25-D for assessment of deficiency associated with renal disease
Test Method	Competitive RIA	Competitive RIA
Detection	^{125}I isotope	^{125}I isotope
Antibody	Polyclonal sheep anti-1,25D	Polyclonal rabbit anti-

Similarities		
Item	**Device**	**Predicate**
	(D_2/D_3)	1,25D (D_2/D_3)
Antigen in calibrators	1,25D (D_3)	1,25D (D_3)
Sample type	Serum or plasma	Serum or plasma
Sample Volume	500 µL	500 µL
Calibration Range	15 – 500 pmol/L (6 – 208 pg/mL)	12 – 480 pmol/L (5 – 200 pg/mL)

Differences		
Item	**Device**	**Predicate**
Assay duration	> 24 hrs	> 2 hrs
Analytical Sensitivity	11 pmol/L (4.6 pg/mL)	5 pmol/L (2 pg/mL)
Reference Range	48 – 150 pmol/L (20.2 – 63.0 pg/mL)	60 – 157 pmol/L (25.1 – 66.1 pg/mL)

K. Standard/Guidance Document Referenced (if applicable):

NCCLS C28-A2 – How to Define and Determine Reference Intervals in the Clinical Laboratory
NCCLS EP9-A – Method Comparison and Bias Estimation Using Patient Samples
NCCLS EP7-P – Interference Testing in Clinical Chemistry
NCCLS EP5-A – Evaluation of Precision Performance of Clinical Chemistry Devices
NCCLS EP6-P2 – Evaluation of the Linearity of Quantitative Analytical Methods

L. Test Principle:

Radioimmunoassay

M. Performance Characteristics (if/when applicable):

1. Analytical performance:
   a. Precision/Reproducibility:
   Precision was evaluated in accordance with NCCLS EP-5A Evaluation of Precision Performance of Clinical Chemistry Devices. Three human serum controls were assayed over 20 assay days spanning more than 90 operating days. The assays were performed by multiple operators using multiple reagents lots.

Control	n	Mean (pmol/L)	Within-run		Total	
			SD	CV%	SD	CV%
A	20	23	2.2	9.7	3.0	13.0
B	20	61	6.5	10.7	6.1	10.1
C	20	122	6.7	5.5	12.0	9.8

Recovery was assayed by adding 1,25D (D_3) to samples prior to extraction and assay. Results are summarized below (1,25D concentrations are in pmol/L).

Sample Concentration	1,25D (D_3) added	Observed Concentration	Recovery (of added analyte)	% Recovery
9.6	38.6	47.4	37.8	98 %
14.8	38.6	55.8	41.1	106 %
84.4	38.6	128.4	44.0	114 %
2.6	38.6	35.5	32.9	85 %
8.6	38.6	40.8	32.2	83 %
59.5	38.6	103.1	43.6	113 %
62.8	38.6	104.4	41.6	107 %
91.7	38.6	129.3	37.6	97 %
			Mean	**101 %**

b. *Linearity/assay reportable range:*
Linearity studies were designed using NCCLS EP6-P Evaluation of the Linearity of Quantitative Analytical Methods. Samples containing varying concentrations of 1,25D prior were assayed in duplicate. The resulting mean concentrations were compared to predicated concentrations. Samples were prepared by diluting a patient sample with assay buffer prior to extraction and assay. The reportable range was determined to be 16 – 253 pmol/L.

c. *Traceability (controls, calibrators, or method):*
Calibrators are assigned values against primary reference calibrators in the Gamma-B 1,25D assay. The mean potency for each calibrator level is calculated from the results of at least 9 independent assays. The assays are carried out on different days by different operators using a variety of reagent batches and expiries.

Acceptable ranges for the controls are assigned by assaying the controls with the Gamma-B 1,25-Dihydroxy Vitamin D assay. A minimum of 20 values are obtained by performing the assay on different days, by different operators using a variety of reagent batches. The value claimed is the mean of the measurements and the acceptable range is given as the mean ± 24%.

Expiration date of 12 months for the calibrators and 18 months for the controls when stored at 4 ºC is based on real time studies that show no or minimal deterioration of the calibrators up to 17 months of storage and up to 25 months for the controls.

Stability of calibrators and controls after they have been reconstituted and stored at -20 ºC, including several freeze thaw cycles, has been established past the recommended 8 week expiry.

d. Detection limit:
The analytical sensitivity of this device was determined to be 11 pmol/L is defined as the concentration corresponding to the mean counts from 10 measurements of the zero calibrator minus 2 standard deviations.

e. Analytical specificity:
Specificity of the antibodies was tested against 2 common metabolites of Vitamin D as well as against the intended analytes. Specificity was determined by comparing the level at which 50% inhibition of ^{125}I binding was reached with that of the intended analyte. The antibody was raised to 1,25D (D_3). Results are summarized below (presented as a percentage of inhibition by 1,25D (D_3)).

Molecule	Cross-reactivity
1,25-Dihydroxyvitamin (D_3)	100.0 %
1,25-Dihydroxyvitamin D_2	91.0 %
24,25-Dihydroxyvitamin D_3	< 0.0123 %
25-Hydroxyvitamin D_3	0.0010 %

Interference testing was conducted in accordance with NCCLS EP7-P. Common endogenous substances – haemoglobin, bilirubin, lipid and urea were tested. No interference was found at the levels tested.

f. Assay cut-off:
Not applicable

2. Comparison studies:
   a. Method comparison with predicate device:
   Accuracy was evaluated based on NCCLS Protocol EP9-A2. The study was conducted by comparing to the marketed device. A comparative study of Gamma-B 1,25-Dihydrox Vitamin D kit and the marketed device on 112 patient samples were performed. Passing & Bablok regression analysis was performed with the following result: IDS = 0.96X – 6.3 (95% confidence intervals of the slope are 0.85 and 1.08 and of the intercept are -15.3 and 1.7) r = 0.82; n = 112.

b. Matrix comparison:
A study comparing serum to plasma (EDTA & Heparin) for Gamma-B 1,25-Dihydrox Vitamin D RIA was performed to establish matrix equivalency. No statistical difference in the means was observed between different sample types.

3. Clinical studies:
   a. Clinical sensitivity:
   Not applicable

   b. Clinical specificity:
   Not applicable

   c. Other clinical supportive data (when a and b are not applicable)
   Not applicable

4. Clinical cut-off:
   Not applicable

5. Expected values/Reference range:
   The 95% reference interval for Normal Adults was determined from 143 apparently healthy adults (13 samples were from the US) and was calculated by a nonparametric method following NCCLS guideline C28-A2, "How to Define and Determine Reference Intervals in the Clinical Laboratory".

   Normal Adults 48 – 150 pmol/L (n=143)
   End-stage of renal disease <11 – 32 pmol/L (n = 44)

   These values are suggested guidelines. It is recommended that each laboratory establish the normal range for the area in which it is located.

N. Conclusion:
The submitted information in this premarket notification is complete and supports a substantial equivalence decision.